JACC Vol. 16, No. 4
October 1990:862–70

Plasma Dehydroepiandrosterone and Dehydroepiandrosterone Sulfate In Patients Undergoing Diagnostic Coronary Angiography

DAVID M. HERRINGTON, MD, MHS,* GARY B. GORDON, MD, PHD, STEPHEN C. ACHUFF, MD, FACC, JORGE F. TREJO, MD, HARLAN F. WEISMAN, MD, FACC, PETER O. KWITEROVICH, MD, JR., THOMAS A. PEARSON, MD, PHD, FACC

Baltimore, Maryland

Serum levels of DHEA sulfate are inversely associated with cardiovascular death in men, and urinary dehydroepiandrosterone (DHEA) levels are inversely associated with clinical manifestations of coronary artery disease. These observations may be related to the antiproliferative effects of DHEA, resulting in inhibition of atherosclerotic intimal hyperplasia. To examine the relation between these steroids and a direct measure of coronary atherosclerosis, plasma DHEA and DHEA sulfate levels were determined in 206 middle-aged patients (103 men, 103 women) undergoing elective coronary angiography.

Plasma DHEA sulfate levels were lower in men with at least one stenosis ≥50% compared with those without any stenosis ≥50% (4.9 ± 2.7 versus 6.1 ± 3.5 nmol/ml, p = 0.05). Levels of DHEA sulfate were also inversely related to the number of diseased coronary vessels (r = −0.20, p = 0.05) and a continuous measure of the extent of coronary atherosclerosis (r = −0.25, p = 0.01) in men. The association between DHEA sulfate levels and extent of coronary artery disease was independent of age and other conventional risk factors for coronary disease. In women, there was no association between plasma DHEA or DHEA sulfate levels and coronary disease.

These data demonstrate a consistent, independent, inverse, dose-response relation between plasma DHEA sulfate levels and angiographically defined coronary atherosclerosis in men. Plasma DHEA sulfate may be another important and potentially modifiable risk factor for the development and progression of coronary atherosclerosis.

(J Am Coll Cardiol 1990;16:862–70)

There is increasing evidence that dehydroepiandrosterone (DHEA) and its sulfate (DHEA sulfate) may play a role in the pathogenesis of coronary artery disease. Although these steroids are major secretory products of the adrenal glands and the primary constituents of urinary and plasma 17-ketosteroids, their role in normal physiology has been difficult to ascertain (1,2). The dramatic age-related decline in urinary (3) and blood (4,5) levels of DHEA and DHEA sulfate that coincides with the increasing incidence of atherosclerosis led several investigators (3,6) to conclude that these steroids have a protective effect against atherosclerosis. Dehydroepiandrosterone has subsequently been shown to be a potent inhibitor of cell growth, proliferation and carcinogenesis (1,2). In cell culture, DHEA inhibits fibroblast growth (7) and differentiation (8). Because atherosclerosis is characterized by differentiation and proliferation of smooth muscle cells and fibroblasts (9), these observations provide a biologically plausible mechanism for a relation between DHEA levels and coronary disease. Recently, in two independently conducted studies (10,11), oral administration of DHEA provided substantial protection against the development of aortic atherosclerosis in cholesterol-fed rabbits.

Several clinical and population-based studies have demonstrated an inverse relation between the level of DHEA or DHEA sulfate, or both, and clinical manifestations of coronary atherosclerosis or its risk factors in humans. Five studies (12–16) have documented low levels of urinary

From The Johns Hopkins University School of Medicine, Department of Medicine, Divisions of Cardiology and General Internal Medicine, the Oncology Center and Department of Pharmacology and Molecular Sciences, and the Lipid Research Atherosclerosis Unit, Department of Pediatrics, The Johns Hopkins Medical Institutions, Baltimore, Maryland. Dr. Herrington was supported by The Andrew W. Mellon Foundation Fellowship in Clinical Epidemiology, New York, New York and Institutional Research Training Grant HL07642 from the National Heart, Lung, and Blood Institute, Bethesda, Maryland. Drs. Gordon and Weisman are recipients of Clinician Scientist Awards from The Johns Hopkins School of Medicine. This work was also supported by PHS Grant P01-CA44530 awarded by the National Cancer Institute and Grant 31497 from the National Heart, Lung, and Blood Institute.

Manuscript received January 10, 1990; revised manuscript received March 7, 1990, accepted April 4, 1990.

*Present address and address for reprints: David M. Herrington, MD, MHS, Division of Cardiology, The Bowman Gray School of Medicine, 300 South Hawthorne Road, Winston-Salem, North Carolina 27103.

0735-1097/90/$3.50

17-ketosteroids or plasma DHEA sulfate in patients who had a prior myocardial infarction (12–15) or a history of heart disease (16) compared with levels in normal control subjects, whereas another study (17) demonstrated the opposite results. One study (12) also identified lower levels of urinary 17-ketosteroids in men with a prior cerebral infarction. Dehydroepiandrosterone or DHEA sulfate levels, or both, have been reported to be inversely associated with cholesterol (18–21), obesity (22,23), diabetes (16) and type A behavior (24). However, other coronary artery disease risk factors such as male gender (4,25–27), hypertension (28–30) and cigarette smoking (16,31,32) have been associated paradoxically with elevated levels of plasma or urinary DHEA sulfate or its metabolites. Barrett-Connor et al. (16,33) reported an independent inverse association between plasma DHEA sulfate levels and the subsequent 12 year cardiovascular and ischemic heart disease mortality rate in a prospective study of 752 men aged 50 to 79 years. In contrast, no association was found between DHEA sulfate levels and cardiovascular or ischemic heart disease mortality rates in a similar cohort of women from the same community (33).

Despite tissue culture, animal model, clinical and population-based data suggesting an important relation between levels of DHEA or DHEA sulfate, or both, and atherosclerosis, there are few data on the relation between these steroids and a direct measure of atherosclerosis in humans. In addition, little is known about the relative importance of plasma levels of DHEA versus DHEA sulfate with respect to atherosclerosis and the apparent disparity between men and women in this relation. The importance of understanding the relation between DHEA and DHEA sulfate levels and coronary atherosclerosis is highlighted by the fact that levels of these steroids may be raised with physical activity (18,23) or oral supplementation (20,34).

This report describes the relation between plasma DHEA and DHEA sulfate levels and angiographically defined coronary atherosclerosis in 206 men and women undergoing coronary angiography. Detailed information is presented on the association between DHEA and DHEA sulfate levels, age, gender and many coronary disease risk factors, as well as the influence these coronary disease risk factors have on the DHEA/coronary atherosclerosis association.

Methods

Study patients. Between April 1985 and April 1988, patients undergoing diagnostic coronary angiography at The Johns Hopkins Hospital were enrolled in The Johns Hopkins Coronary Artery Disease Study to examine the relation between coronary artery disease risk factors and premature coronary atherosclerosis defined by coronary angiography. The study design and informed consent procedure were approved by the Joint Committee on Clinical Investigation of The Johns Hopkins Medical Institutions on May 14, 1985. Patients were eligible for enrollment if they were white men ≤50 years of age or white women ≤60 years of age. Other race groups were not enrolled because anticipated numbers of such participants were too small to permit meaningful race-specific subgroup analyses. Patients were not eligible for enrollment if they lived more than a 3 h drive from Baltimore, were undergoing emergency coronary angiography or percutaneous transluminal coronary angioplasty or had had a myocardial infarction within the previous 12 weeks. Ninety-one percent of the men and 87% of the women who were eligible and contacted concerning the study agreed to participate; 103 men and 103 women were enrolled over the 3 year period.

Data collection. After informed consent was obtained, a standardized questionnaire was administered by a trained interviewer to gather demographic and detailed coronary artery disease risk factor information, determine medication use and measure supine blood pressure at rest, height and weight. The questionnaire included the Rose questionnaire (35), the Framingham type A personality questionnaire (36) and the Paffenbarger physical activity questionnaire (37). The reason for angiography was determined from the catheterization reports for 113 consecutive patients (49 men, 64 women) enrolled after June 13, 1986. Body mass index was calculated as weight (kg)/height (cm^2). On the morning of the procedure or at the time of angiographic examination, a fasting blood specimen was obtained; however, the exact time of day was not recorded. A portion of the plasma was stored at −70°C for subsequent biochemical analysis.

Coronary angiography. Standard coronary angiography was performed, including selective engagement of the right and left coronary arteries and angulated views sufficient to display the coronary tree completely. Coronary angiograms were examined by a consensus panel of three physicians experienced in coronary angiography without knowledge of patients' risk factor status or DHEA and DHEA sulfate levels. On a standardized recording form, each of the 15 American Heart Association (AHA) designated coronary artery segments was characterized as having 0%, 1% to 24%, 25% to 49%, 50% to 74%, 75% to 99% or 100% diameter stenosis and given a corresponding score of 0 to 5, respectively. Bypass grafts, native vessels proximal to bypass grafts and poorly visualized segments were not evaluated. A global measure of the extent of coronary artery disease (coronary disease score) was calculated as the average score for all evaluated AHA segments divided by five. The precision of this coronary disease score was previously established (38) using two blinded evaluations of 55 randomly selected angiograms ($r = 0.96$, $p = 0.001$).

The morphology of coronary lesions was recorded by categorizing each AHA standardized vessel segment as having no disease, a single focal lesion, multiple discrete lesions or diffuse or segmental involvement. A corresponding morphology score of 0 to 3 was assigned to each AHA

segment and a global morphology score was calculated in a fashion similar to the coronary disease score (average AHA segment score divided by three).

Plasma DHEA and DHEA sulfate levels. These were measured using commercially available radioimmunoassay kits (Wien Laboratories and Immuchem). These kits were independently validated with a chromatographic-enzymatic assay (39). The intra- and interassay variabilities of these assays were <10% over a range of values that exceeded those observed in this study. The plasma levels of total cholesterol and triglycerides and the concentrations of low density lipoprotein cholesterol and high density lipoprotein cholesterol were determined using methods of the Lipid Research Clinics Program (40) as modified by Kwiterovich et al. (41).

Statistical analysis. The distributions of DHEA and DHEA sulfate levels were examined using box plots (42). Two men had extremely high values for both DHEA and DHEA sulfate (4 to 6 standard deviations above the mean). One of these men had extensive coronary artery disease and one had trivial coronary disease; both men were heavy smokers. The inclusion of these two patients made no qualitative difference in the associations between DHEA and DHEA sulfate levels and coronary disease; however, the fit of the regression models used to describe the data was markedly improved when they were excluded. The results presented do not include these two extreme cases.

Age adjustment of DHEA and DHEA sulfate levels was performed using least squares estimates of marginal mean values (43), where age was treated as a continuous variable. The distribution of the coronary artery disease and morphology scores deviated significantly from normal; therefore, Spearmans rank order correlation was used to assess the univariate association between DHEA and DHEA sulfate levels and these measures of coronary disease (44). The coronary disease score was also divided roughly into tertiles. All patients with totally normal coronary angiograms and coronary disease scores of zero were assigned to the lowest tertile. The intermediate tertile (0 < coronary disease score < 0.25) identified those patients with mild disease, and coronary disease scores in the highest tertile (coronary disease score > 0.25) were observed in patients with extensive disease.

A polycotomous logistic regression technique was used to examine the relation between DHEA sulfate and coronary disease score tertiles after adjusting for other variables (45). Age was treated as a continuous variable. Odds ratios were calculated from the logistic regression models based on a decrease of 1 standard deviation in DHEA or DHEA sulfate plasma levels. Tests of significance for variables in the logistic regression model were based on the likelihood ratio test.

Table 1. Characteristics of the Study Group According to Gender

	Men	Women
No.	101	103
Median age (yr)	44	52
(range)	(24–50)	(36–60)
Risk factor		
Current smoking (no.)*	81	76
Diabetes mellitus (no.)†	7	12
Mean systolic blood pressure (mm Hg)	119	121
Mean total cholesterol (mg/dl)	225	243
Reason for angiography‡		
Angina or abnormal exercise test, or both (%)	67	72
Atypical chest pain (%)	12	9
Other (%)§	14	6
Missing or undetermined (%)	6	13

*Within last 6 months; †requiring medication; ‡based on catheterization reports from 113 (49 men, 64 women) consecutive patients enrolled after June 13, 1986; §congenital or valvular heart disease, dilated cardiomyopathy or Marfan's syndrome.

Results

Patient characteristics (Table 1). The age difference between men and women was due to the different age entry criteria for men and women. Approximately 75% of the subjects underwent angiography for evaluation of typical angina or a positive exercise tolerance test, or both.

Distribution of DHEA and DHEA sulfate levels according to age and gender (Table 2). In men, there was a steady decline in DHEA and DHEA sulfate levels with increasing age. Linear regression of DHEA and DHEA sulfate levels versus age in men predicted an annual decrease in the DHEA level of 0.53 pmol/ml ($r = -0.41$, $p = 0.0001$) and a yearly decrease in the DHEA sulfate level of 0.21 nmol/ml ($r = -0.35$, $p = 0.0003$). There was no significant association between age and either steroid level in women, although the women with the highest levels were in the youngest age groups.

At ages ≤45 years, men had consistently higher levels of DHEA and DHEA sulfate than women (Table 2). In the 46 to 50 year age group, there was no difference between DHEA levels in men and women, although DHEA sulfate levels were higher in men. Overall, the magnitude of the difference in DHEA and DHEA sulfate levels between men and women was greatest in the younger age groups. After adjustment for age, levels of DHEA sulfate were significantly higher for men than women ($p = 0.002$) and there was a nonsignificant trend toward higher DHEA levels in men.

Association between DHEA and DHEA sulfate levels and presence of coronary artery disease (Tables 3 and 4). Sixty percent of the men and 50% of the women had at least one ≥50% stenosis of a coronary artery as determined by angiography. Dehydroepiandrosterone sulfate levels were signif-

Table 2. Dehydroepiandrosterone (DHEA) and Dehydroepiandrosterone Sulfate (DHEAS) Levels (mean ± SD) in 204 Men and Women Undergoing Diagnostic Coronary Angiography According to Age Group and Gender

Age Group (yr)	Men			Women		
	No.	DHEA (pmol/ml)	DHEAS (nmol/ml)	No.	DHEA (pmol/ml)	DHEAS (nmol/ml)
≤40	18	19.3 ± 5.2*	7.1 ± 3.6	7	11.5 ± 6.7	4.0 ± 1.7
41–45	41	16.0 ± 7.4	5.5 ± 2.7	13	14.6 ± 9.0	2.6 ± 1.5
46–50	42	11.5 ± 4.8	4.5 ± 2.9	22	11.9 ± 6.8	2.9 ± 1.7
51–55				28	11.4 ± 10.0	2.5 ± 2.6
56–60				33	11.4 ± 9.1†	2.6 ± 2.4
All ages	101	14.7 ± 6.7	5.4 ± 3.1	103	11.9 ± 8.7	2.7 ± 2.2
Age adjusted (≤50 yr)		14.4‡	5.2§		13.4‡	3.3§

*Based on 17 subjects; †based on 32 subjects; ‡p = 0.43 for age-adjusted DHEA levels in men versus women (two sample *t* test); §p = 0.0002 for age-adjusted DHEAS levels in men versus women (two sample *t* test).

icantly lower in men with at least one ≥50% stenosis (4.9 ± 2.7 versus 6.1 ± 3.5 nmol/ml, p = 0.05) (Table 3). Dehydroepiandrosterone levels were also lower in men with coronary artery disease defined in this way; however, the difference was not significant compared with men without at least one ≥50% stenosis (14.2 ± 6.8 versus 15.4 ± 6.5 pmol/ml, p = 0.40).

Roughly 40% of the men had totally normal-appearing coronary arteries. Dehydroepiandrosterone sulfate levels were lower in men with any coronary artery disease, regardless of the degree of stenosis, compared with the group with normal-appearing coronary arteries (4.8 ± 2.6 versus 6.7 ± 3.6 nmol/ml, p = 0.01). There was also a nonsignificant trend toward lower DHEA levels in men with any coronary artery disease (14.0 ± 6.7 versus 16.3 ± 6.5 pmol/ml, p = 0.13). In women, there were no differences in DHEA or DHEA sulfate levels between those with and without any stenosis ≥50% or those with totally normal coronary arteries compared with those with any stenosis regardless of the degree of severity.

In men, there was significant, inverse, dose-response relation between DHEA sulfate levels and the number of diseased coronary arteries (any stenosis ≥50%) determined by angiography (Table 4). For DHEA, although mean levels were higher in men with single vessel disease than in those with no disease, there remained an overall statistically significant trend toward lower levels with the increasing number of diseased vessels. In women, there was no association between DHEA or DHEA sulfate levels and the number of diseased vessels.

Association between DHEA and DHEA sulfate levels and coronary artery disease score. Both DHEA and DHEA sulfate levels were inversely associated with the extent of coronary artery disease in men as measured by the coronary disease score (DHEA: r = −0.26, p = 0.01; DHEA sulfate: r = −0.25, p = 0.01). Figure 1 shows the mean levels of DHEA sulfate in men stratified into three age groups and tertiles of the coronary disease score. Within each age group, there was a dose-response relation between the mean level of DHEA sulfate and the extent of coronary disease. After age adjustment, mean DHEA sulfate levels in men with extensive disease (highest coronary disease score tertile) remained significantly lower than levels in men with no disease (lowest coronary disease tertile) (4.9 ± 3.3 versus 6.5 ± 3.0 nmol/ml, p = 0.05). Dehydroepiandrosterone levels in men with extensive disease were only slightly lower

Table 3. Dehydroepiandrosterone (DHEA) and Dehydroepiandrosterone Sulfate (DHEAS) Levels (mean ± SD) in 204 Men and Women According to Presence of Angiographically Defined Coronary Artery Disease* and Gender

CAD	Men (n = 101)			Women (n = 103)		
	No.	DHEA (pmol/ml)	DHEAS (nmol/ml)	No.	DHEA (pmol/ml)	DHEAS (nmol/ml)
+	61	14.2 ± 6.8	4.9 ± 2.7	51	12.1 ± 10.1†	2.7 ± 2.5
−	40	15.4 ± 6.5‡	6.1 ± 3.5	52	11.7 ± 7.1	2.8 ± 1.8
p value§		0.40	0.05		0.83	0.80

*Any ≥50% diameter stenosis; †based on 50 participants; ‡based on 39 participants; §based on two sample *t* test for the presence (+) and absence (−) of coronary artery disease (CAD).

Table 4. Correlation* Between Dehydroepiandrosterone (DHEA) and Dehydroepiandrosterone Sulfate (DHEAS) Levels (mean ± SD) and Number of Diseased Coronary Arteries† in 204 Men and Women According to Gender

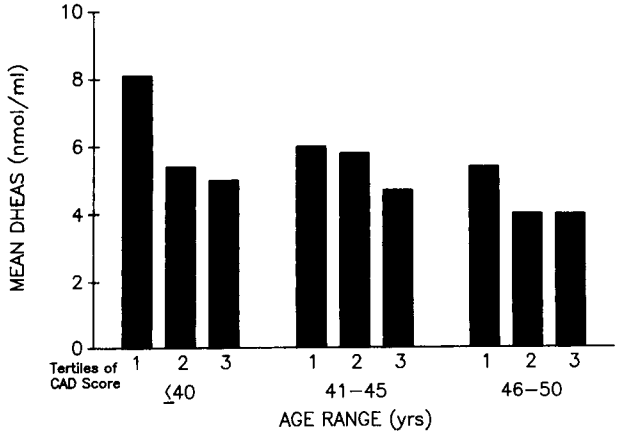
No. of Diseased Coronary Arteries	Men (n = 101)			Women (n = 103)		
	No.	DHEA (pmol/ml)	DHEAS (nmol/ml)	No.	DHEA (pmol/ml)	DHEAS (nmol/ml)
0	40	15.4 ± 6.5‡	6.1 ± 3.5	52	11.7 ± 7.1†	2.8 ± 1.8
1	15	19.0 ± 9.7	5.5 ± 3.5	13	9.8 ± 8.9	1.5 ± 1.0
2	17	14.1 ± 5.8	5.0 ± 2.2	15	9.7 ± 8.1	2.4 ± 2.0
3	29	11.8 ± 3.9	4.4 ± 2.5	23	15.2 ± 11.5§	3.5 ± 3.1
r value		−0.23	−0.20		−0.04	−0.03
p value		0.02	0.05		0.70	0.78

*Spearman rank order correlation coefficient; †≥50% diameter stenosis; ‡based on 39 participants; §based on 22 participants.

than in men with no disease after age adjustment (15.1 ± 7.0 versus 15.7 ± 6.4 pmol/ml, p = 0.77). In women, stratification by age group and coronary disease score tertiles revealed no consistent pattern with respect to DHEA or DHEA sulfate levels.

Association with coronary morphology score, ejection fraction and previous infarction. In addition to the relation to coronary disease score, DHEA sulfate levels were also inversely associated with morphology score in men (r = −0.21, p = 0.06), and men with low DHEA sulfate levels were also more likely to have a low ejection fraction (r = 0.36, p = 0.001). In women, there was no association between DHEA sulfate levels and coronary disease score, morphology score or ejection fraction. In contrast, both men and women with a prior myocardial infarction had lower levels of DHEA sulfate than those without such a history (men: 4.5 ± 2.8 versus 5.7 ± 3.1, p = 0.07; women: 2.1 ± 1.3 versus 3.0 ± 2.4, p = 0.01). Similarly, both men and women with a history of angina had lower levels of DHEA sulfate (men: 5.0 ± 3.1 versus 6.3 ± 4.3, p = 0.09; women: 2.2 ± 1.5 versus 3.0 ± 2.4, p = 0.04).

Figure 1. Mean plasma dehydroepiandrosterone sulfate (DHEAS) levels in 98 men according to extent of angiographically defined coronary atherosclerosis stratified by age group. The age-adjusted mean level in men with no coronary disease (coronary disease [CAD] score tertile = 1) was significantly higher than that in men with extensive coronary disease (coronary disease score tertile = 3) (6.5 ± 3.0 versus 4.9 ± 3.3 nmol/ml, p = 0.05).



Association between DHEA sulfate levels and other coronary artery disease risk factors (Table 5). Dehydroepiandrosterone sulfate levels were 25% to 30% lower in the few men (n = 7) and women (n = 12) with diabetes; however, this difference was not statistically significant. Conversely, being a current smoker and low density lipoprotein cholesterol levels >160 mg/dl were both weakly associated with higher levels of DHEA sulfate in women but not in men. The association between DHEA sulfate levels and alcohol consumption in women was not maintained after adjustment for smoking. There was no significant association between DHEA sulfate levels and systolic or diastolic blood pressure, total cholesterol, high density lipoprotein cholesterol, triglycerides, body mass index or type A personality score in either men or women.

Interestingly, in those men who were not current smokers or diabetic with a systolic blood pressure <160 mm Hg and a total serum cholesterol <240 mg/dl, the association between low DHEA sulfate levels and coronary artery disease (any ≥50% stenosis) was even stronger than in the entire group (4.5 ± 2.6 versus 6.4 ± 3.9 nmol/ml, p = 0.03).

Logistic regression analysis (Table 6). This demonstrated a significant association between DHEA sulfate levels and angiographically defined coronary artery disease in men. After age adjustment, the magnitude of the relative odds of coronary disease was reduced, but there remained a significant age-independent association between DHEA sulfate levels and the relative odds of angiographic coronary disease. Similar trends were observed for DHEA; however, the magnitude and significance of the odds ratios were less than for DHEA sulfate.

To exclude confounding due to other coronary artery

Table 5. Age-Adjusted Dehydroepiandrosterone Sulfate (DHEAS) Levels (nmol/ml) in 204 Men and Women With or Without Coronary Disease Risk Factors

Risk Factor	Men (n = 101)		Women (n = 103)	
	Mean ± SD	p Value*	Mean ± SD	p Value
Current smoker (within last 6 mo.)				
Yes	5.9 ± 2.9	0.43	3.6 ± 2.1	0.03
No	5.3 ± 2.3		2.5 ± 2.2	
Diabetes mellitus (requiring medication)				
Yes	3.8 ± 2.9	0.15	2.1 ± 2.2	0.26
No	5.5 ± 2.9		2.8 ± 2.2	
LDL cholesterol >160 mg/dl				
Yes	5.9 ± 2.9	0.46	3.7 ± 2.2	0.06
No	5.3 ± 2.9		2.6 ± 2.1	
History of hypertension				
Yes	5.3 ± 3.1	0.78	2.9 ± 2.2	0.37
No	5.5 ± 2.9		2.5 ± 2.2	
Family history of early CAD				
Yes	5.7 ± 2.9	0.43	2.9 ± 2.2	0.48
No	5.1 ± 2.9		2.6 ± 2.1	
Physical activity†				
Yes	5.2 ± 2.9	0.46	2.8 ± 2.2	0.65
No	5.6 ± 2.9		2.6 ± 2.2	
Alcohol consumption (within last yr)				
Yes	5.4 ± 2.8	0.93	3.3 ± 2.1	0.006
No	5.3 ± 2.9		2.1 ± 2.1	

*Based on two sample *t* test; †sufficient to work up a sweat ≥ once a week. CAD = coronary artery disease; LDL = low density lipoprotein.

disease risk factors, the age-adjusted associations between DHEA and DHEA sulfate levels and the extent of coronary disease were also examined after adjusting individually for each of the risk factors discussed in the previous section (Table 5). The DHEA sulfate–coronary atherosclerosis relation was also examined after adjusting individually for use of nitrates, digoxin or any cardiac medication. The significant age-adjusted association between DHEA sulfate levels and coronary disease was independent of each of the factors tested (p value range 0.04 to 0.01). Although the magnitude of the age-adjusted odds ratios was smaller for DHEA, it too remained significantly associated with the extent of coronary disease after adjusting individually for each of the factors discussed in the previous section (p value range 0.05 to 0.01) (Table 5). A final model simultaneously adjusting for age, smoking, low density lipoprotein cholesterol and diabetes continued to demonstrate a significant inverse association between DHEA sulfate and the extent of coronary atherosclerosis (p = 0.04).

Table 6. Relative Odds* and Age-Adjusted Relative Odds of Angiographically Defined Coronary Artery Disease for Dehydroepiandrosterone (DHEA) and Dehydroepiandrosterone Sulfate (DHEAS) Levels in 101 Men

			With Age Adjustment	
	RO	p Value†	RO	p Value
DHEA (↓ 6.8 pmol/ml)	1.40	0.08	1.15	0.04
DHEAS (↓ 3.1 nmol/ml)	1.81	0.003	1.32	0.03

*Based on logistic regression models of coronary disease score tertiles versus a decrease of 1 standard deviation for DHEA and DHEAS; †p values derived from the likelihood ratio chi-square for the effect of DHEA or DHEAS. RO = relative odds.

Discussion

DHEA plasma level and extent of coronary artery disease. These data demonstrate an independent, inverse, dose-response relation between plasma dehydroepiandrosterone (DHEA) sulfate levels and the extent of angiographically defined coronary artery disease in men ≤50 years of age undergoing clinically indicated diagnostic coronary angiography. An association was present between DHEA sulfate levels and several different measures of coronary disease including any ≥50% stenosis, any stenosis regardless of severity, number of diseased coronary arteries and a global measure of coronary disease extent (coronary artery disease score). Although the strength of the association between DHEA sulfate levels and the extent of coronary disease was modest (relative odds ratio 1.8), a significant association was maintained after adjustment for age and other coronary disease risk factors (relative odds ratio 1.4). In addition, the

JACC Vol. 16, No. 4
October 1990:862–70

DHEA sulfate level was inversely associated with several angiographic and clinical markers of coronary disease severity, including diffuse lesion morphology, reduced ejection fraction, history of a prior myocardial infarction and angina.

Similar trends were observed between plasma DHEA levels and measures of coronary artery disease in men; however, these trends did not uniformly reach statistical significance. This may seem counterintuitive because DHEA is generally viewed as the active metabolite, and DHEA sulfate is viewed as the chemically stable, storage form of the steroid. However, it is unclear how plasma levels of DHEA sulfate may influence tissue levels of DHEA through the peripherally active sulfatases that can convert DHEA sulfate to DHEA.

Clinically defined coronary artery disease versus coronary atherosclerosis. Previous studies (12–16) have documented a significant association between DHEA sulfate and clinical manifestations of coronary artery disease; however, clinically defined coronary disease involves a variety of pathologic processes in addition to atherosclerotic plaque formation, such as plaque hemorrhage and rupture, thrombosis of stenotic lesions and coronary artery spasm (9). Coronary angiography, unlike clinical diagnosis of coronary disease, is a direct and specific measure of coronary atherosclerosis. The observed association in this study between DHEA sulfate levels and extensive and diffuse coronary disease supports the hypothesis that DHEA sulfate is related to the development of coronary atherosclerotic lesions rather than to the other mechanisms that lead to clinical manifestations of ischemic heart disease.

DHEA and aging. A portion of the association between DHEA sulfate levels and extent of coronary artery disease was accounted for by age. After age adjustment, the odds ratio for coronary disease associated with a 3.1 nmol/ml (1 SD) decrease in the DHEA sulfate level decreased from 1.81 to 1.32. It is unclear whether age-related declines in DHEA and DHEA sulfate levels are simply markers of aging or whether their decline is part of the physiologic mechanism through which aging leads to atherosclerosis. If DHEA sulfate is in the causal chain between aging and atherosclerosis, age adjustment would result in an underestimate of the strength of the association between DHEA sulfate levels and coronary disease. However, even if a decline in plasma DHEA sulfate levels is an epiphenomenon of aging and not in the aging-atherosclerosis causal chain, there remains a significant age-independent association between DHEA sulfate levels and extent of coronary artery disease. Although the subjects in this study were relatively young, the results parallel the findings of Barrett-Connor et al. (16,33) in a cohort of older men and women.

Gender differences in the relation between DHEA sulfate levels and coronary atherosclerosis. In women, no relation between DHEA and DHEA sulfate and angiographic coronary atherosclerosis was found, although women who had had a prior myocardial infarction had lower levels of both steroids than those who had not. If the hypothesis is correct that elevated levels of DHEA or DHEA sulfate, or both, inhibit atherosclerosis, it is difficult to explain why women have lower levels of plasma and urinary DHEA and DHEA sulfate and yet have less atherosclerosis than men. One possible explanation is that endogenous estrogen or some other female-specific attribute attenuates the risk of a low DHEA or DHEA sulfate level in women. Alternatively, the beneficial effect of high levels of DHEA or DHEA sulfate in men may not be due to an antiproliferative effect, but rather to aromatization of DHEA to estrogen in peripheral tissues. In this case, women who normally have high levels of circulating endogenous estrogen would be unlikely to derive a large additional benefit from conversion of high levels of DHEA to estrogen.

DHEA and coronary artery disease risk factors. The observed trend toward higher levels of DHEA sulfate in smokers, especially in women, is consistent with previous reports (16,31,32) and points out the complex relation among these steroids, coronary disease risk factors and coronary atherosclerosis. The small number of men and women with diabetes had age-adjusted DHEA sulfate levels that were 25% to 30% lower than those without diabetes; however, these differences did not reach statistical significance. Dehydroepiandrosterone inhibits fat synthesis (8) and increases tissue insulin sensitivity in mice (46); however, its role in human diabetes has not been well defined. It is not known whether low levels of plasma DHEA or DHEA sulfate, or both, contribute to or are the result of diabetes; however, it is possible that low levels of these steroids in diabetes is one of the mechanisms through which diabetes is associated with coronary artery disease.

Limitations of the current study. There are two major limitations of this study. First, these are cross-sectional data; it is possible that a low DHEA sulfate level is a consequence rather than a cause of coronary atherosclerosis. However, the consistency of results from this study with other prospective population-based data (16) and the antiatherogenic effect of DHEA administration in an animal model of atherosclerosis (10,11) suggest that low plasma levels of DHEA sulfate are an antecedent to coronary atherosclerosis. In addition, the similarity of the association among several different measures of coronary artery disease, the dose-response relation observed and the fact that the association between DHEA sulfate and coronary disease was independent of age and other risk factors support the hypothesis that DHEA sulfate is causally related to the pathogenesis of coronary atherosclerosis.

The second limitation is that the study group is a highly select one. Those undergoing coronary angiography are not representative of the population at large with respect to traditional coronary artery disease risk factors or symptoms consistent with coronary disease (47). Because clinicians do

not check DHEA sulfate levels before deciding to refer a patient for angiography, a direct referral bias with respect to DHEA sulfate levels is unlikely. Furthermore, the DHEA sulfate–coronary disease association was not confounded by any of the coronary disease risk factors tested or the presence or absence of angina as defined by the Rose questionnaire (35). Nevertheless, confounding due to other unidentified factors remains a possibility.

Patients undergoing elective angiography also are not representative of all patients with coronary atherosclerosis. Patients with severe disease resulting in sudden death or unstable angina requiring emergency angiography were not studied; however, exclusion of patients with severe disease would make it more difficult to detect an association between DHEA or DHEA sulfate levels and coronary atherosclerosis if the associations hold in these subsets of patients as well. In general, caution should be used in applying these results to the general population or assuming that the findings pertain to all forms of coronary atherosclerosis.

Implications for future investigation of coronary atherosclerosis. These data raise the question of whether modifying DHEA sulfate levels could alter the risk of the development and progression of atherosclerosis. Some studies suggest that a low calorie diet (48) or physical activity (18,23) can result in higher levels of plasma DHEA or DHEA sulfate. Furthermore, DHEA sulfate levels can be elevated through oral administration. Two studies have shown that DHEA (20) or DHEA sulfate (34) can be orally administered for up to 42 weeks in normal subjects, resulting in significant increases in plasma DHEA sulfate levels without apparent adverse side effects. These observations suggest that oral DHEA could be used to elevate plasma DHEA sulfate levels safely. However, more observational data, animal models and information about the bioavailability, pharmacokinetics and potential side effects of orally administered DHEA are needed before testing the hypothesis that such an intervention alters the natural history of coronary artery disease in humans.

We are indebted to the entire staff of the Lipid and Lipoprotein Analysis Laboratory at The Johns Hopkins Hospital. Special thanks go to Hazel Smith for her invaluable role in recruitment, data collection and lipid analysis and to Carol Derby, PhD for her essential contribution in interviewing patients and data management. We also wish to thank Paul Talalay, MD, Myron Weisfeldt, MD and Paul Whelton, MD for their helpful comments and suggestions.

References

1. Gordon GB, Shantz LM, Talalay P. Modulation of growth, differentiation and carcinogenesis by dehydroepiandrosterone. Adv Enzyme Regul 1987;26:355–82.
2. Schwartz AG, Whitcomb JM, Nyce JW, Lewbart ML, Pashko LL. Dehydroepiandrosterone and structural analogs: a new class of cancer chemopreventive agents. Adv Cancer Res 1988;51:391–424.
3. Marmorston J, Griffith GC, Geller PJ, Fishman EI, Wlesch F, Weiner JM. Urinary steroids in the measurement of aging and of atherosclerosis. J Am Geriatr Soc 1975;23:481–92.
4. Migeon CJ, Keller AR, Lawrence B, Shepard TH. Dehydroepiandrosterone and andosterone levels in human plasma: effect of age, sex, day-to-day and diurnal variation. J Clin Endocrinol Metab 1957;17:1051–62.
5. Orentreich N, Brind JL, Rizer RL, Vogelman JH. Age changes and sex differences in serum dehydroepiandrosterone sulfate concentrations throughout adulthood. J Clin Endocrinol Metab 1984;59:551–5.
6. Kask E. 17-Ketosteroids and arteriosclerosis. Angiology 1959;10:358–68.
7. Saenger P, New M. Inhibitory action of dehydroepiandrosterone (DHEA) on fibroblast growth. Experientia 1977;33:966–7.
8. Gordon GB, Newitt JA, Shantz LM, Weng DE, Talalay P. Inhibition of the conversion of 3T3 fibroblast clones to adipocytes by dehydroepiandrosterone and related anticarcinogenic steroids. Cancer Res 1986;46:3389–95.
9. Schwartz SM, Ross R. Cellular proliferation in atherosclerosis and hypertension. Prog Cardiovasc Dis 1984;26:355–72.
10. Gordon GB, Bush DE, Weisman HF. Reduction of atherosclerosis by administration of dehydroepiandrosterone: a study in the hypercholesterolemic New Zealand white rabbit with aortic intimal injury. J Clin Invest 1988;82:712–20.
11. Arad Y, Badimon JJ, Badimon L, Hembree WC, Ginsberg HN. Dehydroepiandrosterone feeding prevents aortic fatty streak formation and cholesterol accumulation in cholesterol-fed rabbit. Arteriosclerosis 1989;9:159–66.
12. Marmorston J, Lewis JJ, Bernstein JL, et al. Excretion of urinary steroids by men and women with myocardial infarction. Geriatrics 1957;12:297–300.
13. Weiner JM, Marmorston J. Statistical techniques of difference. Ann N Y Acad Sci 1969;161:641–69.
14. Rao LGS. Urinary excretion patterns after acute myocardial infarction. Lancet 1970;2:390–1.
15. Lopez-S A. Metabolic and endocrine factors in aging. In: Rothschild H, Chapman CF, eds. Risk Factors for Senility. New York: Oxford University Press, 1984:210–9.
16. Barrett-Connor E, Khaw K-T, Yen SSC. A prospective study of dehydroepiandrosterone sulfate, mortality, and cardiovascular disease. N Engl J Med 1986;315:1519–24.
17. Zumoff B, Troxler RG, O'Connor J, et al. Abnormal hormone levels in men with coronary artery disease. Arteriosclerosis 1982;2:58–67.
18. Lopez-S A, Wingo C, Hebert JA. Total serum cholesterol and urinary dehydroepiandrosterone in humans. Atherosclerosis 1976;24:471–81.
19. Sonka J, Fassati M, Fassati P, Gregorova I, Picek K. Serum lipids and dehydroepiandrosterone secretion in normal subjects. J Lipid Res 1968;9:769–72.
20. Adlercreutz H, Kerstell J, Schauman KO, Svanborg A, Vihko R. Plasma lipids and steroid hormones in patients with hypercholesterolaemia or hyperlipaemia during dehydroepiandrosterone sulphate administration. Eur J Clin Invest 1972;2:91–5.
21. Biozel R, de Peretti E, Cathiard AM, et al. Pattern of plasma levels of cortisol, dehydroepiandrosterone and pregnenolone sulphate in normal subjects and in patients with homozygous familial hypercholesteroleaemia during ACTH infusion. Clin Endocrinol 1986;25:363–71.
22. Lopez-S A, Krehl WA. A possible interrelation between glucose-6-phosphate dehydrogenase and dehydroepiandrosterone in obesity. Lancet 1967;2:485–7.
23. Sonka J, Gregorova I, Tomsova Z, et al. Plasma androsterone, dehydroepiandrosterone and 11-hydroxycorticoids in obesity: effects of diet and physical activity. Steroids Lipids Res 1972;3:65–74.
24. Fava M, Littman A, Halperin P. Neuroendocrine correlates of the type A behavior pattern: a review and new hypotheses. Int J Psychiatry Med 1987;17:289–307.

25. De Peretti E, Forest MG. Pattern of plasma dehydroepiandrosterone sulfate levels in humans from birth to adulthood: evidence for testicular production. J Clin Endocrinol Metab 1978;47:572–7.

26. Yamaji T, Ibayashi H. Plasma dehydroepiandrosterone sulfate in normal and pathological conditions. J Clin Endocrinol Metab 1969;29:273–8.

27. Zumoff B, Bradlow HL. Sex difference in the metabolism of dehydroisoandrosterone sulfate. J Clin Endocrinol Metab 1980;51:334–6.

28. Sekihara H, Osawa N, Kosaka K. Serum dehydroepiandrosterone sulfate and dehydroepiandrosterone levels in essential hypertension. J Clin Endocrinol Metab 1975;40:156–7.

29. Shao A, Nowaczynski W, Kuchel O, Genest J. Secretion rate of dehydroepiandrosterone sulfate in benign essential hypertension as compared to normal subjects. Can J Biochem 1970;48:1308–13.

30. Nowaczynski W, Fragachan F, Silah J, Millette B, Genest J. Further evidence of altered adrenocortical function in hypertension: dehydroepiandrosterone excretion rate. Can J Biochem 1968;46:1031–8.

31. Dai WS, Gutai JP, Kuller LH, Cauley JA, for the MRFIT Research Group. Cigarette smoking and serum sex hormones in men. Am J Epidemiol 1988;128:796–805.

32. Khaw K-T, Tazuke S, Barrett-Connor E. Cigarette smoking and levels of adrenal androgens in postmenopausal women. N Engl J Med 1988;318:1705–9.

33. Barrett-Connor E, Khaw K-T. Absence of an inverse relation of dehydroepiandrosterone sulfate with cardiovascular mortality in postmenopausal women (letter). N Engl J Med 1987;317:711.

34. Nestler JE, Barlascini CO, Clore JN, Blackard WG. Dehydroepiandrosterone reduces serum low density lipoprotein levels and body fat but does not alter insulin sensitivity in normal men. J Clin Endocrinol Metab 1988;66:57–61.

35. Rose G, McCartney P, Reid DD. Self-administration of a questionnaire on chest pain and intermittent claudication. Br J Prevent Social Med 1977;31:42–8.

36. Haynes SG, Levine S, Scotch N, Feinleib M, Kannel WB. The relationship of psychosocial factors to coronary heart disease in the Framingham Study. I. Methods and risk factors. Am J Epidemiol 1978;107:362–83.

37. Wilson PW, Paffenbarger RS Jr, Morris JN, Havlik RJ. Assessment methods for physical activity and physical fitness in population studies: a report of a NHLBI workshop. Am Heart J 1986;111:1177–92.

38. Pearson TA. Risk Factors for Arteriographically Defined Coronary Artery Disease (PhD dissertation). Baltimore: The Johns Hopkins School of Hygiene and Public Health, Department of Epidemiology, 1983.

39. Holtzclaw WD, Gordon GB. Measurement of serum levels of dehydroepiandrosterone sulfate: a comparison of radioimmunoassay and enzymatic analysis. Steroids 1989;54:355–71.

40. Manual of Laboratory Operations: LRC Program 1: Lipid and Lipoprotein Analysis. US Dept of Health, Education, and Welfare publication no. NIH 75-628, May 1974 (revised version issued October 1982).

41. Kwiterovich PO Jr, White S, Forte T, Bachorick PS, Smith H, Sneiderman A. Hyperapobetalipoproteinemia in a kindred with familial combined hyperlipidemia and familial hypercholesterolemia. Atherosclerosis 1987;7:211–25.

42. Tukey JW. Exploratory Data Analysis. Reading, MA: Addison-Wesley, 1977:39–41.

43. Wilcosky TC, Chambless LE. A comparison of direct adjustment and regression adjustment of epidemiologic measures. J Chron Dis 1985;38:849–56.

44. Snedecor GW, Cochran WG. Statistical Methods. 7th ed. Ames, IA: Iowa State University Press, 1980:192.

45. Walker SH, Duncan DB. Estimation of the probability of an event as a function of several independent variables. Biometrika 1967;67:167–79.

46. Coleman DL, Leiter EH, Schwizer RW. Therapeutic effect of dehydroepiandrosterone (DHEA) in diabetic mice. Diabetes 1982;31:830–3.

47. Kottke BA, Zinsmeister AR, Holmes DR Jr, Kneller RW, Hallaway BJ, Mao SJT. Apolipoproteins and coronary artery disease. Mayo Clin Proc 1986;61:313–20.

48. Hendrix A, Heyns W, DeMoor P. Influence of low-calorie diet and fasting on the metabolism of dehydroepiandrosterone sulfate in obese subjects. J Clin Endocrinol 1968;28:1525–33.